Update

Cellulosic Ethanol — Tomorrow's Sustainable Energy Source

Rita D'Aquino

This past January, President Bush spelled out the terms of a new energy policy during his State of the Union Address that set an ambitious goal for the nation to displace 20% displacement of fossil fuels with renewable or alternative fuels by 2017. In terms of capacity, this translates as 35 billion gal/yr of renewable fuel that must be in use in 10 years — nearly five times the 2012 target now in place.

This plan may initially appear unattainable, considering that in the U.S. alone, the total market for automotive fuels is currently 140 billion gal/yr. Of this amount, 5–6 billion gal/yr of production capacity, or less than 5%, is currently met by ethanol that is primarily derived from corn and other grains. Corn-based ethanol production is expected to plateau at 12 to 15 billion gallons gal/yr — not quite near what is required to meet the President's goal of producing 35 billion gal/yr of renewable fuels by 2017.

Consuming more fiber

Against this backdrop, cellulosic biomass, including wood, cereal straws and grasses has emerged as a viable, sustainable source for liquid transportation fuels, and shows promise in comprising a major portion of the 20% in Bush's "20-in-10" goal. Some industry experts say cellulosic ethanol is the closest we will get to a petroleum substitute that will sustain our future energy needs. A case in point is a study conducted in 2005 by the National Renewable Energy Laboratory (NREL; www.nrel.gov) with the U.S. Dept. of Agriculture (USDA; www.usda.gov) showing that the U.S. could produce enough raw biomass (forest residues, fast-growing trees and switchgrass) — the major constituent of which is cellulose fiber — to equal about 60% of all the oil that is used in the U.S. The problem is, unlike corn, this raw biomass is difficult to convert. A primary deterrent for economic utilization of cellulose has been the high cost of enzymes used in the hydrolysis of cellulose to produce the sugars that are fermented into ethanol.

Over the past several years, however, process development efforts have been directed at economically producing cellulase enzymes at a lower cost. According to Thomas Nagy, president of Novozymes North America (www.novozymes.com), the firm is strongly concentrating its R&D efforts on cellulosic ethanol. Funded by a U.S. Dept. of Energy (DOE; www.doe.gov) back in 2000, research conducted by the firm has culminated in a reduction in the cost of conversion [of cellulose into fermentable sugars] by a factor of 30, notes Nagy. Meanwhile, Genencor (www.genencor.com) partnering with NREL for the past four years, has reached similar results — an estimated cellulase cost in the range of $0.10-$0.20/gal of ethanol — approximately a 30-fold improvement in enzyme economics in NREL's cost model.

"We believe the dawn of cellulosic ethanol is close at hand," says Jack Huttner, vice president, business development for biomass of Genencor. The firm's Stargen line of granular starch hydrolyzing enzymes, for example, offers the ethanol industry greater yields with fewer processing steps, while requiring less energy, materials and capital. Previously, ethanol plants cooked grains and other starchy feedstocks with thermostable enzymes to begin the process of converting starch to fermentable sugars. The Stargen enzymes include blends of an alpha amylase and a glucoamylase that convert granular or uncooked starch to sugars on a continuous basis through a simultaneous saccharification and fermentation process — without the need for a cook step.

Last December, Genencor announced an agreement to supply enzymes to a new biomass-to-ethanol demonstration facility that Mascoma Corp. (www.mascoma.com) plans to build in Rochester, NY, in the next 10–12 months. The facility is expected to utilize a number of New York State agricultural and/or forest products as

DuPont, among other firms, are developing ways to produce ethanol from such feedstocks as corn stover. Photo courtesy of Pioneer Hi-Bred International.

biomass, including paper sludge, wood chips, switch grass and corn stover. Genencor is also part of a research consortium sponsored the French National Research Agency (www.ademe.fr) to develop a conversion system that will add ethanol production to paper pulp mills.

Integrated biorefineries

As part of its strategy to accelerate biofuels production by enabling the adoption of efficient, high-performance, bio-based technologies, DuPont (www.dupont.com) has teamed up with the DOE to develop a fermentation process that allows high conversion of both C-6 glucose sugars and the difficult-to-ferment C-5 xylose sugars to ethanol at high yields. This integrated biorefinery technology, which uses a microorganism called *Zymomonas mobilis* to make the conversions, was recently licensed to ethanol producer Broin, Inc. (www.broin.com). Broin is currently transforming its Voyager Ethanol site, located in Emmetsburg, IA, from a 50-million gal/yr corn dry-mill facility into a 125-million gal/y biorefinery that integrates corn fractionation and lignocellulosic conversion technologies for the production of ethanol from corn grain and corn stover, respectively.

"The integrated biorefinery technology will significantly increase the amount of ethanol per acre achievable by using both the corn grain and stover," explains DuPont Biofuels vice president and general, manager John Ranieri. This facility is expected to produce 11% more ethanol from a bushel of corn and 27% more ethanol from an acre of corn while using 83% less energy than what is needed to operate a corn to ethanol plant.

Meanwhile, Diversa (www.diversa.com) is developing enzyme cocktails to convert different types of cellulosic biomass into fermentable sugars as part of an overall objective of developing a new, more cost-effective process. In 2005, as the first key step toward this goal, it developed a set of candidate enzymes under the Integrated Corn-Based Biorefinery (ICBR) program led by DuPont, a U.S. Department of Energy-sponsored consortium to develop an economical, commercial-scale process to convert starch and cellulosic biomass to fuel ethanol and other value-added chemicals. A change in just a single amino acid can greatly affect the function of a protein such as an enzyme or an antibody, notes Diversa, which heralds a suite of patented, state-of-the-art gene evolution technologies, called DirectEvolution. Comprising Gene Site Saturation Mutagenesis (GSSM) and Tunable GeneReassembly (TGR) technologies, DirectEvolution technologies provides potentially significant competitive advantages, including the ability to generate the broadest amount of genetic sequence diversity, the ability to make fine changes across an entire gene, and the freedom to use unrelated genes when recombining starting genes.

For example, genes that were used for development of Valley "Ultra-Thin", a novel alpha amylase enzyme optimized for the cost-effective production of ethanol from corn, were recovered from an organism that thrived under conditions similar to those required for starch liquefaction for ethanol production. Through a combination of sequence-based and functional screening, three amylase candidate enzymes were selected from the organism, each of which exhibited one of the optimal characteristics for the performance specification: optimal activity at pH 4.5, optimal thermal stability at 105°C, and optimal expression in the selected production host. The three genes encoding the candidate enzymes were combined using Diversa's DirectEvolution platform, and a variant that possessed the optimal characteristics of all three parental genes was selected for product development. The resulting "super alpha amylase," known as Ultra-Thin, outperforms competitive enzymes and is currently marketed by Valley Research for use in starch liquefaction for the production of ethanol.

Last month, Diversa took a bold step to enhance its enzyme business with an aggressive push into cellulosic ethanol plant development and production. The firm signed a definitive merger agreement with biofuel process developer Celunol (www.celunol.com) to create "the first company possessing fully integrated technologies for cellulosic ethanol production," — *i.e.,* the first to offer integrated end-to-end capabilities in pre-treatment, novel enzyme development, fermentation, engineering, and project development. Headquartered in Cambridge, MA, the new entity, 76% of which is owned by Diversa, hopes to be recognized globally as a producer of cellulosic ethanol and as a strategic partner in biorefineries around the world. The combined company plans to bring its first U.S. commercial-scale cellulosic ethanol plants into production by late 2009.

Meanwhile, Celunol officially broke ground last month on a new demonstration-scale cellulosic ethanol production facility, located at its Jennings, LA, site, that will use locally grown sugarcane bagasse and specially-bred energy cane to produce ethanol. The plant is slated for completion before the end of 2007 with a design capacity of 1.4 million gal/yr of ethanol. It will feature Celunol's patented technology, which uses a combination of microorganisms and specialty enzymes to convert up to 95% of available sugars in biomass feedstocks into biofuel product. Celunol also recently finalized the construction of a 50,000-gal/yr pilot cellulosic ethanol facility. Said to be the first of its kind in the U.S., the facility will further refine technology for the conversion of biomass such as sugarcane bagasse, wood chips and other abundant biomass sources, into ethanol. The company has also licensed its technology to Tokyo-based Marubeni Corp. (www.marubeni.com) and has incorporated the process into BioEthanol Japan's 1.4 million L/yr cellulosic ethanol plant in Osaka, Japan.

Mimicking a methodology that Diversa made famous, Lucigen Corp. (www.lucigen.com) has scoured the hot springs of Yellowstone National Park to find thermophiles that produce especially hearty enzymes for cellulosic digestion in fermentation processes. Lucigen relies on a grant from the U.S. Department of Energy's Joint Genome Institute to identify the genetic structure of the organisms Lucigen has isolated. We mine the genome for (enzymes) we're interested in," says company president and founder David Mead, who developed a cloning technique [for genes of interest] that allows duplication of very small amounts of DNA, "which is important because at extremely high temperatures — of about 185°F — bacteria levels are extremely low," Mead says.

Lucigen has come up with a promising enzyme that helps break down the fibers in corn byproducts used to make ethanol, so that more fuel can come from the same amount of corn. Nicknamed CornBuster 1, and the subject of a patent application, the enzyme functions well in the heat of alcohol production, effecting an increase in the amount of alcohol produced from corn by 2–3%, according to Mead. "That may not sound like much, but if a plant makes 50 million gallons of corn-alcohol a year, that's 1 million extra gallons a year, or $2 million in revenue," he says. Field trials are underway and Lucigen will likely apply for Environmental Protection Agency approval later this year to sell the CornBuster 1 commercially.

BIOBUTANOL ON THE HORIZON

In the rush to develop and promote corn-derived bioethanol, the U.S. might be ignoring another viable biofuel — biobutanol. It possesses many attractive properties as a fuel. For example, its energy content is closer to gasoline than that of ethanol's, and its low vapor pressure facilitates its use in existing gasoline supply and distribution channels. Compared to ethanol, biobutanol is less volatile, not sensitive to water, less hazardous to handle, less flammable, has a slightly higher octane number, and can be mixed with gasoline in any proportion. However, its high production costs — resulting in an average cost of $3.75/gal — have prevented its widespread use as a fuel.

This scenario is changing. Existing bioethanol capacity can be cost-effectively retrofitted to biobutanol production with minor changes in fermentation and distillation procedures. In fact, DuPont and BP joined forces (www.bpdupontbiofuels.com) and are working with British Sugar, a subsidiary of Associated British Foods plc, to produce commercial quantities of biobutanol at a Wissington, U.K.-based facility, which is scheduled to go online this year. It will involve the conversion of British Sugar's bioethanol plant to facilitate the output of 30,000 m.t./yr of biobutanol. To expedite commercial market entry, initial biobutanol production will be based on an existing technology that uses local supplies of corn, sugar cane or beets as feedstock.

In a similar effort, Oxfordshire, U.K., biotechnology firm, Green Biologics (GBL; www.greenbiologics.com) has recently demonstrated the use of cellulosic biomass as a feedstock for its biobutanol product, Butafuel. At the start of this year, the firm received $1.1 million in funding to support the development of Butafuel. The Dept. of Trade and Industry-led technology program is providing $494,000, and shareholder investors and business owners are providing the rest.

GBL has isolated a cocktail of thermophilic microorganisms that enable the rapid enzymatic hydrolysis and release of fermentable sugars from biomass. The company plans to integrate this patented hydrolysis technology with a proprietary butanol fermentation process and is partnering with EKB Technology (www.ekbtechnology.gbr.cc) to improve yields and further lower Butafuel production costs. "Our expertise in microbial strain development, together with EKB's innovative use of non-edible food stocks, should lead to a step change in the economic viability of the manufacturing process for butanol. With our Butafuel technology, we are aiming for a two to three fold reduction in cost," says Ed Green, president and founder of GBL. Patents are pending on the Butafuel process, while GBL moves the technology from a research to development stage.

The road ahead

Significant progress has been made in proving enzymes could be a cost-effective technology in producing ethanol from cellulosic materials. "But, we're not there yet. There is still a lot of hard work left to do in the pre-treatment area, in enzymatic hydrolysis, and in research on the fermentation side," said Michael Pacheco, director of the National Bioenergy Center (NBC) at NREL, at a panel discussion during President Bush's visit to Novozymes' U.S. headquarters (Franklin, NC) on February 22, 2007. "One of the things that we're trying to do is to develop transitions that grow from existing industries. We have to begin the use of a broader base of fuels [such as switchgrass, a native grass to the Midwest that grows in relatively poor conditions and can survive droughts very well] and grow crops in areas where we're not currently growing anything," he continues. "If we grow crops that are specifically designed to produce feedstocks that are adapted well for the enzymes that Novozymes is producing, it would really increase ethanol yields," Pacheco exclaimed.

DOE is currently considering proposals to work with industry in the area of developing fermentation organisms, "which is a really important part, because the yeast — the bugs today that we use to make ethanol — won't work in the future with these new feedstocks," said Pacheco. CEP